

# OBSERVATIONS ON THE MORPHOLOGY AND LIFE-HISTORY OF MICROPHALLUS LIMULI N. SP. (TREMATODA: MICROPHALLIDAE)

# HORACE W. STUNKARD

# Department of Biology, New York University, University Heights, New York City

During the summer of 1950, Dr. T. H. Waterman, while studying the finer structure of the eyes of the horseshoe crab, *Limulus polyphemus*, observed small cysts, each of which contained a larval worm. Cysts containing living larvae were referred to me for study and possible identification. The wall of the cyst is very tough and it was impossible by mechanical means to liberate the larvae without injury. Some of the cysts were left in sea water and two days later two of them had ruptured, freeing the larvae which were alive and active. The worms were almost sexually mature ; the testes and seminal vesicles contained motile spermatozoa. The excretory system was traced completely and the morphology of the metacercariae showed them to be a species of *Microphallus*. An abstract of the findings was presented at the summer meeting of the Marine Biological Laboratory (Stunkard, 1950).

Twenty-eight specimens of *L. polyphemus*, collected in Pleasant Bay, Orleans, Mass., and with a size-range from 5 cm. carapace length to fully grown individuals, were examined and all were infected. Cysts were present in the connective tissue of the digestive gland and other organs as well as in the eyes, and in all of them the larvae were apparently representatives of a single species.

Cysts were fed to baby chicks, half-grown white mice and young golden hamsters. One chick was examined 2 days and another 5 days after feeding, but no worms were found. Examination of the birds' feces did not yield eggs of the parasite and birds killed later than 5 days after feeding were free of infection. One mouse died 12–16 hours after the first feeding and many excysted metacercariae were found in the initial portion of the small intestine. Another mouse, killed 2 days after feeding, contained worms with eggs in their uteri and all of 6 mice examined 2-4 days after feeding contained mature worms. After 5 days the number of worms diminished and none were found after 9 days. No worms were found after subsequent feedings of metacercariae, indicating a possible immunity from the first infection. The feeding of cysts to golden hamsters gave results substantially like those with mice. From these feeding experiments and the rapid elimination of the worms, it appears that mice and hamsters are not natural or normal hosts of the parasite. However, the recovery of sexually mature specimens makes it possible to describe the species and compare it with other related ones. The specimens can not be assigned with certainty to any known species and accordingly they are described as a new species for which the name Microphallus limuli is proposed. Specimens must be designated in some way and since it is simpler and easier to drop a name in syonymy than to distinguish between different species which have been erroneously included under a single name, no serious inconvenience will result if the present specimens should eventually prove to belong to a previously described species.

## DESCRIPTION

Cysts (Fig. 1) removed from *L. polyphemus* were oval, colorless, 0.16-0.2 mm. in length and 0.12-0.18 mm. in width. The wall was 0.009-0.011 mm. in thickness, hyaline, and resolvable into two layers, approximately equal in thickness. This wall was probably produced by material from cystogenous cells of the cercaria. It was enclosed in a thin, colorless, fibrous coat, probably deposited as a reaction product by the host.

Released from the cyst a metacercaria, under moderate coverglass pressure, measured 0.24–0.40 mm, in length and 0.10–0.20 mm, in width. Fixed, stained



FIGURE 1. A; metacercaria in cyst, from L. *polyphemus*. B; outline drawing of fixed and stained specimen, 0.314 mm. long, with 84 eggs in the uterus, to show characteristic shape when elongate.

and mounted, this larva measured 0.30 by 0.19 mm. In it the testes, ovary and vitellaria had attained almost complete development; the testes and seminal vesicle contained motile spermatozoa. The size and location of the organs are shown in Figure 2. The flame cell formula is 2[(2+2) + (2+2)], which so far as known is characteristic of all members of the family Microphallidae. The cuticula bears imbricate, flattened spines which become progressively smaller posteriorly and may be absent behind the acetabulum. In living specimens the edges of the body tend to bend ventrally, forming a rim around an elongate depression. Typically there is a slight constriction at the level of the bifurcation of the digestive tract, and the body may become pyriform with either the anterior or posterior portion wider than the other. In front of the digestive ceca the body is filled with minute, unicellular glands.

Sexually mature worms (Figs. 1B; 4) from mice and hamsters are very little, if any, larger than the metacercariae. The suckers and individual organs are no larger in adult worms than in the larger metacercariae. The only increase in size

results from a bulbous expansion of the posterior one-third to one-half of the body, which may be almost spherical and when bent ventrally produces a claviform appearance. The increase in size of the posterior region is caused by the development of the uterus and the accumulation of eggs in it. More than 100 eggs have been counted in a single worm. The increase in thickness and massing of eggs obscure the organs in the posterior portion of the body and they are more conspicuous in the metacercaria.



FIGURE 2. Fixed and stained metacercaria, 0.32 mm. long, ventral view, flattened to show excretory and genital structures.

Adult specimens, when alive and under moderate coverglass pressure, measure 0.26–0.40 mm. in length and 0.15–0.20 mm. in greatest width. The terminal egg-filled portion of the body is always wider and thicker than the flatter anterior region; otherwise the shape is unchanged from that in the metacercaria. The acetabulum measures 0.029–0.035 mm. in diameter and is located about one-third of the body length from the posterior end, a little farther forward relatively than in the metacercaria. The mouth is subterminal; the oral sucker is 0.035–0.045 mm. in diameter; the prepharynx is 0.01–0.033 mm. in length; the pharynx is 0.017–0.021 mm. in

diameter; the esophagus is 0.1–0.13 mm. in length; the intestinal crura, which are preacetabular, diverge at a wide angle and measure 0.08–0.10 mm. in length. Fixed and stained specimens are somewhat smaller; the smallest gravid worm is 0.26 mm. long; 0.11 mm. in greatest width; in diameter the oral sucker is 0.025 mm., the acetabulum is 0.0225 mm., and the pharynx is 0.012 mm. The uterus of this worm contains 14 eggs.

The excretory system is unchanged from that in the metacercaria. The pore is terminal; there is a preporal sphincter, a bicornuate bladder, from which the collecting ducts pass anteriad as shown in Figure 2.

The testes are lateral, opposite, situated at least partially in the acetabular zone and about midway between the anterior and posterior ends of the enlarged portion of the body; they are oval to ovate, 0.05–0.065 mm. in the transverse axis and 0.03–



FIGURE 3. Composite drawing of two adjacent, slightly oblique, 10  $\mu$  sections, to show relations of acetabulum, gonads, genital pore, male papilla, vitelline ducts (one of which is enlarging before joining with the one from the opposite side), and dorsal to it the initial coil of the uterus containing a newly formed egg; ac, acetabulum; ed, ejaculatory duct; gp, genital pore; iu, initial coil of uterus; mp, male papilla; ov, ovary; sv, seminal vesicle; ts, testis; ut, uterus; vd, vitelline duct. Parenchyma not shown.

0.04 mm. in the longitudinal one. Sperm ducts arise from the median faces, pass mediad and anteriad, join above the acetabulum, and the common duct expands into a large, transverse, oval seminal vesicle which may be as large as one of the testes and is located medially or slightly to the left in the area directly behind the digestive ceca. From the vesicle, an ejaculatory duct curves posteriad, mediad, and ventrad. It terminates in the male papilla which opens into the genital atrium near the left margin of the acetabulum (Fig. 3). The distal portion of the seminal vesicle and the ejaculatory duct are enclosed in glandular cells. The male papilla is very small, 0.006–0.0075 mm. in diameter and 0.012–0.014 mm. long.

The ovary is oval to triangular, longer transversely, 0.06–0.065 by 0.036–0.045 mm., situated in the area bounded by the right cecum, the right testis, the acetabulum and the seminal vesicle. The details of the female reproductive ducts were observed in serial sections. The oviduct arises from the postero-median face of the ovary and

#### MICROPHALLUS LIMULI NEW SPECIES

coils posteriad and ventrad; there is a small enlargement which contains spermatozoa and from which Laurer's canal winds dorsad to open near the midline, just behind the level of the acetabulum. Shortly after the origin of Laurer's canal, the oviduct turns dorsad and anteriad, receives a short common vitelline duct, and expands into the ootype. Mehlis' gland consists of only a few cells. The vitellaria are large, lobed glands, situated on either side, filling the regions between the testes and the posterior wall of the body. Ducts from the two sides pass mediad and anteriad; they unite to form the common vitelline duct which opens into the oviduct as noted



FIGURE 4. Fixed and stained specimen, 0.295 mm. long, with 120 eggs in the uterus, dorsal view, flattened to show details of the reproductive organs; dc, digestive cecum; os, oral sucker; ov, ovary; ph, pharynx; sv, seminal vesicle; ts, testis; vt, vitellaria.

above. The initial portion of the uterus is dorsal to the acetabulum (Fig. 3). The uterus passes posteriad in a sinuous course near the median plane almost to the end of the body, then coils anteriad, ventrally, along the right side of the body to the level of the cecum, turns posteriad and passes backward mediad to the ascending limb; near the posterior end of the body the uterus crosses to the left side of the body where it forms a loop similar to the one on the right side and the recurrent limb turns mediad to open into the genital atrium. The course of the uterus becomes more coiled and irregular as it is filled with eggs. The eggs are oval to ovate, operculate, 0.016–0.02 by 0.009–0.011 mm., and some of them have a very small knob at the antopercular end. Eggs in the initial portion of the uterus are almost colorless; those on the left side of the body are bright yellow. Mature eggs contain ciliated miracidia. In the metacercaria, the vitelline follicles form a compact triangular mass, extending on each side from the testis to the posterior end of the body; in gravid specimens the vitellaria and testes are enclosed by coils of the uterus, which in part obscure their outlines.

Second intermediate host: Limulus polyphemus.

Definitive hosts: experimentally, white mice and golden hamsters.

First intermediate host : unknown.

Type and paratype specimens : deposited in the Helminthological Collection of the U. S. National Museum, slide No. 47592.

## Discussion

The taxonomic position of the species described in the present paper is obscured by certain peculiar difficulties. The asexual stages, and the natural hosts of the adult are yet unknown. Development in mice and hamsters may have produced specimens substantially different from those maturing in normal hosts. The present specimens undoubtedly belong to the genus Microphallus, but the generic concept is still somewhat tentative and many of the specific descriptions are incomplete. The genus Microphallus was erected by Ward (1901) to contain a species described by him (1894) as Distoma opacum. The worms were found in fresh-water fishes: Amia calva, Ictalurus punctatus, and Perca flavescens from Lake St. Clair, Michigan. Metacercarial stages were encysted in crayfish, Cambarus propinguus. Stossich (1899) erected the genus *Levinsenia* and included *D. opacum* in it, but Looss (1899) argued that D. opacum Ward did not belong in Levinschia and that probably it represented a genus as yet unnamed. Ward (1901) defined the new genus Micro*phallus* and proposed a new subfamily, Microphallinae, to contain it and *Levin*seniella. The subfamily Microphallinae was included in the family Heterophyidae Odhner, 1914 by several subsequent authors, including Fuhrmann (1928), although Travassos (1921) had removed the subfamily from the Heterophyidae and elevated it to family status. The more recent morphological and life-history studies of Rankin (1939a, 1939b, 1940a, 1940b), of Cable and Hunniuen (1940), Baer (1943) and other authors have established the Microphallidae as a family distinct from Heterophyidae.

As noted, Stossich (1899) erected the genus Levinscuia to contain four species: Distomum brachysomum Creplin, 1837; D. macrophallus v. Linstow, 1875; D. pygmaeum Levinsen, 1881; and D. opacum Ward, 1894. In the same year, Lühe and Looss, independently, designated L. brachysoma as type of Levinsenia. The next year, Jägerskiöld (1900) proposed L. pygmaeum as type of the genus. Ward (1901) designated D. opacum as type of a new genus Microphallus; he stated that Levinsenia Stossich was preoccupied (Levinsenia Mesnil, 1897) and that Stiles and Hassall were to propose the name Levinseniella to replace it. Although the paper by Stiles and Hassall did not appear until 1902, the name Levinseniella was validated by Ward (1901) with Levinseniella brachysoma as type species. Jägerskiöld (1901), although aware of the announcement by Ward, proposed the name Spelotrema for the invalid name Levinsenia, with S. pygmaeum as type. He stated, p. 982, "Ich sehe aber aus Ward's Aufsatz: On the Structure of the Copulatory Organs of *Microphallus* n. g. (Studies from the Zoological Labor. The Univ. of Nebraska), dass Stiles mir zuvorgekommen ist, und lasse daher, obgleich ich den Aufsatz von Stiles noch nicht habe finden können, seinen Namen *Levinseniella* gelten." Jägerskiöld (1904) listed *Levinseniclla* as a synonym of *Spelotrema* although he (1907) recognized the two as distinct genera. Jägerskiöld (1901) proposed *Spelotrema* as a replacement for *Levinsenia* and regarded the two as identical, but actually this is not the case since *L. brachysoma* was designated as type of *Levinseniella* and *L. pygmaenm* as type of *Spelotrema*, and the two species are not congeneric. The name *Spelotrema* has had an unsavory reputation and the history of the name and of the generic concept were reviewed by Cable and Hunninen (1940). They discussed the nomenclatorial problem outlined above. However, the distinctness of the species allocated to *Spelotrema* and those included in *Levinseniella* was demonstrated by the studies of Rankin previously cited.

The problem of *Spelotrema* appears to be solved. Baer (1943) stated that there are no morphological differences which distinguish *Spelotrema* from *Microphallus*, and that the two genera had been maintained because species of *Spelotrema* are typically parasites of birds whereas those of *Microphallus* occur in fishes and amphibians. Baer argued that difference in host is not a proper criterion for generic distinction, since the metacercariae of these species are encysted in crustaceans which are eaten by both birds and fishes. The metacercariae are well developed and become sexually mature in a few days after ingestion by the definitive hosts. Moreover, certain species mature in mammals as well as birds and host-specificity is not at all precise. Since *Microphallus* antedates *Spelotrema*, the latter name was suppressed as a synonym and the species formerly included in *Spelotrema* has been accepted by Rausch (1947), Rausch and Locker (1951) and by Cable and Kuns (1951).

The number of valid species in the genus Microphallus is yet uncertain. Osborn (1919) described M. ovatus from Micropterus dolomieu taken in Lake Chautauqua, New York. Strandine (1943) compared specimens of Microphallus from Amia calva and Micropterus dolomicu of Lake Lelanau, Michigan, and found no significant difference between M. opacus and M. ovatus. He reported overlapping variations in total size, size of acetabulum, extent of spination, length of ceca, and size of eggs in the two previously accepted species. Rausch (1946a, 1946b) discussed morphological variation and host-specificity of species in the genus Microphallus and reported natural infections by M. ovatus in three species of turtles and in the racoon. He (1947) found that microphallid cysts from crayfish fell into two distinct size groups, the larger of which (1.3 by 0.9 mm.) agrees with the measurements given by Ward (1894) for cysts of M. opacus. Although the cysts were of two sizes, even from the same cravfish, the metacercariae were reported to be morphologically identical and all were able to mature when fed to suitable experimental hosts. He was unable to except the larvae by mechanical means, but found that they emerged in 48 hours when placed in Ringer's solution with fragments of the gland in which they were found. On release, they began to copulate and when left in normal saline, Ringer's solution, or media devised for culture of protozoa, they matured and produced large numbers of apparently normal eggs. He fed metacercariae to various kinds of possible hosts; the cysts failed to hatch in chicks

and toads; the metacercariae emerged but were voided in an immature condition by certain snakes. Sexually mature worms were recovered from two species of turtles, two snakes, the opossum and the racoon, but only negative results were obtained with salamanders, frogs, chicks, albino rats and a skunk. Adults from the two types of cysts differed in size, but since they were otherwise so similar, Rausch concluded that they belong to a single species. Furthermore, he suggested that M. gracilis Baer, 1943, may also be identical with M. opacus. These experiments of Rausch are of fundamental significance, since they exemplify the surest method of determining specific relations among parasitic worms. His data suggest that there are actually two species represented in his material, and M. ovatus may be distinct from M. opacus.

Diagnoses of previously described species in the genus Microphallus (syn. Spelotrema) were made by Rankin (1940a), Cable and Hunninen (1940), Baer (1943) and Rausch and Locker (1951). Earlier authors, Odhner (1905), Nicoll (1907), and more recent ones, Rankin, Cable and Hunninen, and Baer, regarded the size of the male papilla as a diagnostic, specific feature, whereas Rausch and Locker emphasized the length of the digestive ceca and the relative size of suckers. Rankin (1940a) described the life-cycle of Spelotrema papillorobusta n. sp., and listed the specific details of five previously described species. Cable and Hunninen (1940) tabulated the specific features of five species which they compared with Spelotrema *nicolli*, a species described by them as new and for which they traced the life-history. Baer (1943) described M. gracilis n. sp., from Neomys fodiens, suppressed Spelotrema as a synonym of *Microphallus*, and recognized fourteen species, thirteen of which were distinguished in a key. The fourteenth species, M. minus Ochi, 1928, was not included as the description was published in Japanese. The genus Monocaecum Stafford, 1903, was suppressed as a synonym of *Microphallus* and its only species, M, baryurus was listed as possibly identical with M, ovatus. Baer noted that the metacercariae attain almost complete maturity in their crustacean intermediate hosts, that they become gravid in a few hours in the anterior portion of the intestine of various cold-blooded and warm-blooded hosts, and that they diminish in numbers from day to day as they migrate toward the posterior portion of the intestine. To account for these facts, he suggested that the metacercariae have developed a "progenèse retardée" or "néoténie naissante," and that the cold-blooded hosts may be "hôtes d'attente" to which the parasites have become adapted. Rausch and Locker (1951) described M. enhydrae n. sp., from the sea-otter, Enhydra lutris, and recognized fourteen species in the genus *Microphallus*.

The present specimens agree most closely with the description of *M. claviformis* (Brandes, 1888), described originally from the rectum of *Tringa alpina* at Halle, Germany. Brandes stated that the body was often constricted into two regions, with a flattened anterior portion about twice as long as a swollen posterior portion, which gave the worms a club-shape and suggested the specific name, *Distomum claviforme*. The description and figure give an incomplete, inadequate, and probably erroneous characterization of the species. Lühe (1899) stated, "Bei *Dist. claviforme* Brds., welches sich durch die auffällige Länge von Praepharynx und Oesophagus auszeichnet, ist die Lage und Form der Dotterstöcke und die Lage des Genitalporus vollkommen unbekannt, so dass die Art meines Erachtens zur Zeit in ein System nicht eingereiht werden kann." Jägerskiöld (1900) added, "Was aber *Distomum claviforme* Brandes betrifft, so bin ich, mit der Ansicht Lühe's anschliessend, eher

geneigt zu glauben, dass es zu den nie zu identifizierenden Arten gehört, wenn es nicht etwa dem Autor selbst gelingen wird, es wiederzufinden." Jägerskiöld suspected, and justifiably, that Brandes had misidentified the internal organization, that the reported anterior testis was the seminal vesicle, that the posterior testis was the cirrus sac or gonotyl, and that the actual testes and vitellaria were obscured by the windings of the uterus. Brandes suggested that the worms might be the adult stage of a metacercaria encysted in the crab, *Carcinus maenas*.

What was presumed to be the same species was redescribed by Nicoll (1907) as *Spelotrema claviforme* from *Pelidna* (*Tringa*) alpina and *Aegialis histicula*. He noted that the sexually mature worms were smaller than the metacercariae encysted in *C. maenas*, thus nullifying the suggestion of Brandes. In a second paper, Nicoll (1909) restated the specific diagnosis, included a figure, and added *Anthus obscurus*, *Numenius arquata*, *Moticilla flava* and *Larus ridibundus* as new hosts of the parasite. Apparently Brandes and Nicoll are the only authors who have published original observations of the species, although it had been transferred to *Brachycoelium* by Stossich (1892) and to *Lecithodendrium* by Stossich (1899).

The only morphological feature in which the present specimens differ from the description of Nicoll is in the size of the male papilla, which is only about one-half the size of that in the specimens described by Nicoll. The worms from *Limulus* may indeed be specifically identical with those described by Nicoll, and at first they were referred to M. claviformis. But bionomic features seem to preclude the allocation of the specimens to that species. In all other microphallids for which the life-history is known, the metacercariae occur in crustaceans. While larvae of this species may encyst in crustaceans, the intense infection in Linulus suggests that the horseshoe crab is the natural host, and Limulus polyphemus, a living representative of the Xiphisurida, is a chelicerate form related more closely to the pycnogonids and the scorpions than to the crustaceans. It is far removed, zoologically and phylogenetically, from the Crustacea. Moreover, *Limulus* does not occur on the coast of Europe and some other animal must serve there as the second intermediate host of M. claviformis. Adult stages of the present species have so far been recovered only from mammals; and possible bird hosts, other than the chick, have not been tested. If the species is actually M. claviformis, it should mature in Larus ridibundus when young birds are available. Presumably, older and previously infected birds would prove refractory.

Other species of *Microphallus* reported from mammalian hosts include *M. minus* by Ochi (1928) from rats, cats, dogs and man in Japan; *M. brevicaeca* by Africa and Garcia (1935) from man in the Philippine Islands; *M. gracilis* by Baer (1943) from *Neomys fodiens* taken in the environs of Geneva, Switzerland; *M. opacus* by Rausch (1947) from the opossum and the racoon; and *M. enhydrae* by Rausch and Locker (1951) from the arctic sea-otter. The metacercariae of *M. minus* and *M. opacus* occur in crustaceans and *M. opacus* matured in both poikilothermal and homothermal hosts. Since Rausch has shown that *M. opacus* can mature in opossums and racoons, the fact that the present specimens developed in mammals may be incidental and insignificant. *M. opacus*, like the present species, failed to infect chicks. It is entirely probable that the metacercariae of the species are discovered, it will be possible to attempt infection of crustaceans and learn whether or not the parasite can use both arachnids and crustaceans as second intermediate hosts. Since the microphallid cercariae are very immature and the metacercariae undergo extensive development

in the second intermediate host, it must be more than a mere transfer host. In the present state of uncertainty concerning specificity in the genus *Microphallus* it seems inappropriate to assign the specimens described in this paper to *M. claviformis*, and they are designated as *M. limuli* n. sp. Actually the features employed by previous authors to distinguish between species of *Microphallus* are exceedingly variable and tenuous; and certain of the presently accepted species are probably identical. The experimental results of Rausch (1947) have thrown doubt on the validity of several species and, in correspondence, Cable has recently suggested that *M. nicolli* may be the same as *M. simile*.

In the paper mentioned earlier, Baer (1943) divided the family Microphallidae Travassos, 1921, into two families, Microphallidae and Maritrematidae, distinguished by the absence of a cirrus sac in the former and the presence of this organ in the latter. In the family Microphallidae, as restricted, he included the genera: Microphallus Ward, 1901; Levinseniella Stiles and Hassall in Ward, 1901; Spelotrema Jägerskiöld, 1901; Monocaecum Stafford, 1903; and Spelophallus Jägerskiöld. 1908. The family Maritrematidae, as conceived, contained the genera: Maritrema Nicoll, 1907; Microphalloides Yoshida, 1938; Gynaecotyla Yamaguti, 1939 (misspelled Gynaecocotyla,= Cornucopula Rankin, 1939); and Pseudospelotrema Yamaguti, 1939 (= Maritreminoides Rankin, 1939). It is to be noted that although Baer included five genera in the restricted family Microphallidae, in the same paper he suppressed Spelotrema and Monocaecum as identical with Microphallus. Moreover, he noted that *Spelophallus* differs from *Spelotrema* in only a single feature; that in the former genus the metraterm opens into the distal part of the genital atrium whereas in the latter it opens deeper in the atrium, near the base of the male papilla. Spelophallus is known by only a single species, S. primus Jägerskiöld, 1908. In these small, soft-bodied worms, where lack of skeletal support and contraction of different sets of muscles can produce extensive variation in the relations of such mobile parts, it is questionable whether Spelophallus has any true morphological basis. In my opinion, the differences described by Jägerskiöld can have no more than specific value. Accordingly, Spelophallus is suppressed as a generic concept and the species, S. primus, is transferred to Microphallus as M. primus (Jägerskiöld, 1908).

The proposal of Baer to divide the family Microphallidae was analyzed by Cable and Kuns (1951). They erected a new genus, *Carneophallus*, to contain *C. trilobatus* n. sp., and recognized a series of genera from *Microphalloides* and *Pseudospelotrema* to *Levinseniclla* and *Carneophallus*, showing gradual reduction of the cirrus sac correlated with increased complexity in structure of the copulatory organs. Accordingly, they rejected the proposal of Baer, retained the genera with a cirrus sac in the family Microphallidae, and presented a scheme to illustrate probable evolution in the family. The argument of Cable and Kuns seems convincing and there appears to be no adequate basis for recognition of a family Maritrematidae.

#### SUMMARY

Metacercariae from *Limulus polyphemus* developed to sexual maturity in white mice and golden hamsters. The worms are described as a new species, *Microphallus limuli*. Morphologically they are very similar to *M. claviformis* (Brandes, 1888), but bionomic features seem to preclude their allocation to that species. The genus

Microphallus is discussed, Spelophallus Jägerskiöld, 1908 is suppressed as a synonym and S. primus Jägerskiöld, 1908, the only known species, is transferred to Microphallus as M. primus (Jägerskiöld, 1908). In agreement with Cable and Kuns (1951), the family Maritrematidae Baer, 1943 is not accepted.

### LITERATURE CITED

- AFRICA, C. M., AND E. Y. GARCIA, 1935. Heterophyid trematodes of man and dog in the Philippines with descriptions of three new species. Philip. Jour. Sci., 57: 253-267.
- BAER, JEAN G., 1943. Les trématodes parasites de la musaraigne d'eau Neomys fodiens (Schreb.). Bull. Soc. Neuchât. Sci. Nat., 68: 34-84.

BRANDES, G., 1888. Helminthologisches. Arch. Naturg., Jg. 54: 247-251.

- CABLE, R. M., AND A. V. HUNNINEN, 1940. Studies on the life history of Spelotrema nicolli (Trematoda: Microphallidae) with the description of a new microphallid cercaria. Biol. Bull. 78: 136-157.
- CABLE, R. M., AND M. L. KUNS, 1951. The trematode family Microphalidae with the description of Carncophallus trilobatus n. g., n. sp., from Mexico. Jour. Parasitol., 37: 507-514.
- FUHRMANN, O., 1928. Trematoda. In: Kükenthal-Krumbach, Handbuch der Zoologie; Bd. II, Hälfte 1: 140 pp.
- JÄGERSKIÖLD, L. A., 1900. Levinsenia (Distomum) pygmaca Levinsen, ein genitalnapftragendes Distomum. Centralb. Bakt. Parasit., Orig. 27: 732–740. JÄGERSKIÖLD, L. A., 1901. Tocotrema expansum (Crepl.) (= Monostomum expansum Crepl.)
- eine genitalnapftragende Distomide. Centralb. Bakt. Parasit., Orig. 30: 979-983.
- JÄGERSKIÖLD, L. A., 1904. Scaphanoccphalus expansus (Crepl.) eine genitalnapftragende Distomide. Rcs. Swed. Zoöl. Exped. Egypt, part 1: 1-16.
- JÄGERSKIÖLD, L. A., 1907. Zur Kenntnis der Trematoden-gattung Levinseniella. Zool. Stud. Till. Tullberg, Uppsala, pp. 133-154.
- JÄGERSKIÖLD, L. A., 1908. Kleine Beiträge zur Kenntnis der Vogeltrematoden. Centralb. Bakt. Parasit., Orig. 48: 302-317.
- Looss, A., 1899. Weitere Beiträge zur Kenntnis der Trematoden-Fauna Acgyptens. Zool. Jahrb., Syst., 5: 521-784.
- LÜHE, M., 1899. Zur Kenntnis einiger Distomen. Zool. Anz., 22: 524-539.
- NICOLL, W. A., 1907. Observations on the trematode parasites of British birds. Ann. Mag. Nat. Hist., ser. 7, 20: 245-271.
- NICOLL, W. A., 1909. Studies on the structure and classification of the digenetic trematodes. Quart. Jour. Micros. Sci., n. s., 53: 391-487.
- OCHI, S., 1928. On a new trematode, Microphallus minus n. sp., having Macrobrachium nipponensis as its intermediate host. Tokyo Iji Shsh., No. 2578, pp. 4-12. Original in Japanese; English abstract by K. Morishita in Jap. Jour. Zool., 2 (2): 1.
- ODHNER, T., 1905. Die Trematoden des arktischen Gebietes. Fauna Arctica, 4: 291-372.
- OSBORN, H. L., 1919. Observations on Microphallus ovatus sp. nov. from the crayfish and black bass of Lake Chautauqua, N. Y. Jour. Parasitol., 5: 123-127.
- RANKIN, JOHN S., JR., 1939a. Studies on the trematode family Microphallidae Travassos, 1921. I. The genus Levinschiella Stiles and Hassall, 1901, and description of a new genus, Cornucopula. Trans. Amer. Micros. Soc., 58: 431-447.
- RANKIN, JOHN S., JR., 1939b. Studies on the trematode family Microphallidae Travassos, 1921. III. The genus Maritrema Nicoll, 1907, with description of a new species and new genus, Maritreminoides. Amer. Midland Natur., 22: 438-451.
- RANKIN, JOHN S., JR., 1940a. Studies on the trematode family Microphallidae Travassos, 1921. II. The genus Spelotrema Jägerskiöld, 1901, and description of a new species, Spelotrema papillorobusta. Trans. Amer. Micros. Soc., 59: 38-47.
- RANKIN, JOHN S., JR., 1940b. Studies on the trematode family Microphallidae Travassos, 1921. IV. The life cycle and ecology of Gynaecotyla nassicola (Cable and Hunninen, 1938) Yamaguti, 1939. Biol. Bull., 79: 439-451.
- RAUSCH, R., 1946a. New host records for Microphallus ovatus Osborn, 1919. Jour. Parasitol., 32: 93-94.

- RAUSCH, R., 1946b. The racoon as a new host for *Microphallus* sp. with additional notes on *M. ovatus* from turtles. *Jour. Parasitol.*, **32**: 208–209.
- RAUSCH, R., 1947. Some observations on the host relationships of *Microphallus opacus* (Ward, 1894) (Trematoda: Microphallidae). *Trans. Amer. Micros. Soc.*, **66**: 59–63.
- RAUSCH, R., AND BETTY LOCKER, 1951. Studies on the helminth fauna of Alaska. II. On some helminths parasitic in the sea otter, *Enhydra lutris* (L.). Proc. Helminth. Soc. Wash. D. C., 18: 77-81.
- Stossicii, M., 1892. I distomi degli uccelli. Lavoro monografico. Boll. Soc. adriat. sci. nat., Trieste, 13: 143-196.
- STOSSICH, M., 1899. Los membramento dei Brachycoclium. Boll. Soc. adriat. sci. nat., Trieste, 19:7-10.
- STRANDINE, ELDON' J., 1943. Variation in *Microphallus*, a genus of trematodes, from fishes of Lake Lelanau, Michigan. *Trans. Amer. Micros. Soc.*, 62: 293-300.
- STUNKARD, H. W., 1950. Microphallid metacercariae encysted in *Limulus polyphemus*. *Biol. Bull.*, **99**: 347.
- TRAVASSOS, L., 1921. Contribuição para conhecimento o da fauna helmintolojica brasileira—IX. Sobre as especias da sub-familia Microfallinae Ward, 1901. Arch. Esc. Sup. Agr. Mcd. Vct., 4: 84–91.
- WARD, H. B., 1894. On the parasites of the lake fish. I. Notes on the structure and life history of *Distoma opacum*, n. sp. *Proc. Amer. Micros. Soc.*, **15**: 173–182.
- WARD, H. B., 1901. Notes on the parasites of the lake fish. III. On the structure of the copulatory organs in *Microphallus*, nov. gen. *Trans. Amer. Micros. Soc.*, 22: 175–187. Reprinted (1901) in *Stud. Zool. Lab.* Univ. Nebraska.